Vulnerability of tropical shellfishes against PSP contamination during bloom of *Pyrodinium bahamense* var. *compressum*

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Abstract—Accumulation of paralytic shellfish poisoning (PSP) toxins in representative shellfish samples of epifaunal, partially exposed benthic and infaunal nature in association with the toxic dinoflagellate, *Pyrodinium bahamense* var. *compressum* was investigated. Shellfish and seawater samples were collected during the last quarter of 2007 and in 2008 in Sorsogon Bay, Philippines. All tissue samples were then analyzed for PSP toxins while cell density of *P. bahamense* var. *compressum* was determined in water samples. Results showed that epifaunal, partially-exposed benthic, and infaunal shellfish have remarkable differences in terms of PSP toxin accumulation. Epifaunal shellfish can accumulate PSP toxin of more than a thousand times compared to infaunal shellfish with the latter negative for PSP toxin during *P. bahamense* var. *compressum* bloom.

Key words: Paralytic shellfish poisoning, *Pyrodinium bahamense* var. *compressum*, epi-fuanal shellfish, in-faunal shellfish, partially exposed benthic shellfish

Introduction

Paralytic shellfish poisoning (PSP) toxins contamination in shellfish has become an important concern in the Philippines due to the increasing number of coastal areas affected by toxic bloom of dinoflagellate Pyrodinium bahamense var. compressum (Hermes and Villoso 1983, Estudillo and Gonzales, 1984 Bajarias et al. 2006, Relox Jr. et al. 2009). Related to these blooms are reported poisoning cases implicating several species of bivalve mollusks (Furio and Gonzales 2002, Relox et al. 2009). In response, the Philippines Government employs continuous monitoring program that plays a key role in implementing management strategies (Gonzales et al. 1989). Part of the strategies used is the enforcement of closures for areas affected by PSP encompassing all bivalves to ensure protection of public health (Bajarias et al. 2006). However, the limited data on PSP toxin accumulation to support such strict regulations policy on shellfish draws some criticism from fishery sectors because of detrimental effects to the shellfish industry.

Previously, PSP toxin accumulation was confirmed in tropical shellfish but limited to six partially-exposed benthic and epifaunal species (Montojo et al. 2006), as well as one infaunal species (Montojo et al. 2010) of bivalves. PSP toxin

accumulation in several shellfish species from the tropics have not yet established despite prevalence of toxic bloom.

In this study, PSP toxin accumulation of several epifaunal, partially-exposed benthic, and infaunal shellfish species collected from the bay was evaluated and compared during bloom of *P. bahamense* var. *compressum*.

Materials and Methods

Seasonal shellfish toxicity

Five to ten specimens of each shellfish species were collected monthly in Sorsogon Bay, southeastern Luzon Is., Philippines (Fig. 1) during the last quarter of 2007 and in 2008. Specimens of infaunal shellfishes embedded 20–30 cm were collected from different types of habitat in the Bay, *Lingula* sp., venus clams, *Katelysia* spp. from sandy-muddy tidal flats and *Batissa violacea* from mangrove area. Partially-exposed benthic species like *Atrina pectinata*, *Scapharca globosa*, *S. indica*, *Azorinus* sp., *Pharella* sp., *Placuna placenta* and *Pugilina* sp., were also collected in the area. Epifaunal species like *Perna viridis* and *Crassostrea* spp. were also collected as reference species for toxin accumulation

After shellfish specimens were thoroughly cleaned and

the total length was measured, whole tissue removed from shells was weighed and extracted for toxin analysis using AOAC (1990) method. Toxins in the extracts were analyzed by High Performance Liquid Chromatography (HPLC) according to Oshima (1995a). Toxicity of each shellfish was expressed as μg STX-eq. $100^{-1} g^{-1}$ tissue, which was calculated from nmol g^{-1} obtained by HPLC analysis using specific toxicity values of each toxin component (Oshima 1995a). Individual toxicity was also expressed as μg STXeq/specimen following similar calculation method above.

Seasonal abundance of P. bahamense var. compressum

During shellfish collection, water samples were also collected at surface and bottom layers by 2.51 Niskin bottle (General Oceanics, Canada) and fixed with formalin. A 500 ml of fixed water sample was concentrated to 20 ml by passing through a sieve with a mesh size of $20 \,\mu\text{m}$. Pyrodinium bahamense var. compressum cell in 1 ml of concen-



Fig. 1. The sampling area.

trated water was counted under light microscope using Sedgwick-Rafter chamber. The cell density of P bahamense var. compressum was expressed as cells l^{-1} of seawater.

Results

Toxin accumulation in shellfish in relation with abundance of P. bahamense var. compressum cells

Figure 2 shows the seasonal toxicity of shellfishes (2A–B) in relation with the abundance of *P. bahamense* var. compressum (Fig. 2C-D). Both partially-exposed benthic and epifaunal species became positive of PSP toxins parallel with the occurrence of P. bahamense var. compressum. Considerable toxicity of more than the international regulation limit of $80 \,\mu g$ STXeq. $100^{-1} \, g^{-1}$ tissue was recorded in *Pharella* sp. $(274 \,\mu\mathrm{g}\ \mathrm{STXeq}.\ 100^{-1}\,\mathrm{g}^{-1})$, Crassostrea sp. $(628 \,\mu\mathrm{g}\ \mathrm{STXeq}.$ $100^{-1} \,\mathrm{g}^{-1}$), A. pectinata (1,047 µg STXeq. $100^{-1} \,\mathrm{g}^{-1}$), S. globosa $(1,129 \,\mu g \, STXeq. \, 100^{-1} \, g^{-1})$, P. placenta $(1,488 \,\mu g \, I)$ STXeq. $100^{-1} \,\mathrm{g}^{-1}$) and P. viridis (22,616 $\mu\mathrm{g}$ STXeq. $100^{-1} \,\mathrm{g}^{-1}$) during peak of P. bahamense var. compressum bloom indicating that these species are not fit for human consumption. Azorinus sp. and S. indica accumulated minimal level of toxicity at 12 and 75 μ g STXeq. 100^{-1} g⁻¹, respectively.

On the contrary, three species of infaunal shellfishes, *K. hyantina*, *B. violacea*, *Lingula* sp. and a partially-exposed benthic univalve species, *Pugilina* sp., showed negative of PSP toxins during the entire study period.

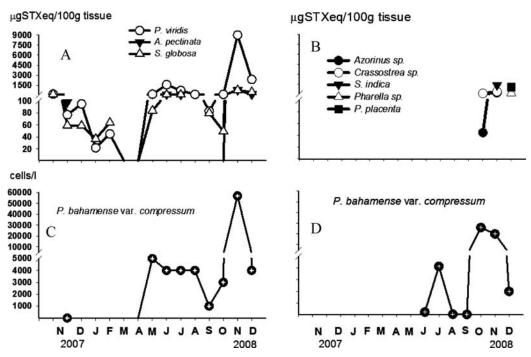


Fig. 2. Seasonal toxicity of shellfish (A, B) and the occurrence of P. bahamnese var. compressum (C, D) in Sorsogon Bay.

Discussion

Sorsogon Bay is a source of different shellfish species of varying commercial value ranging from local consumption to both local and international markets. Perna viridis, being a sentinel species is commonly implicated for paralytic shellfish poisoning events during bloom of Pyrodinium bahamense var. compressum, thus this study was conducted to investigate the vulnerability of the various shellfish resources to PSP. Results showed that not all the bivalve species became toxic during bloom of P. bahamense var. compressum. While bivalve species like P. viridis had toxin level much greater than the tolerable limit according to the national bulletins from 2006 to 2008 (BFAR, unpublished), infaunal species' PSP toxicity level remained consistently negative or less than that of the tolerable limit. Results are in conjunction with previous study of Montojo et al. 2010, in which Paphia undulata an infaunal species was focused on.

It has been shown in several studies that epifaunal and partially-exposed benthic species accumulate PSP toxin. One of these studies reported on temperate epifaunal species like scallops, oysters and mussels, and partially-exposed benthic species like short necked clam that showed PSP toxin accumulation when exposed to Alexandrium tamarense both in the wild and fed in rearing tanks (Sekiguchi et al. 2001). Another is the work of Montojo et al. (2006) that showed similar observation in tropical waters of Masinloc Bay for epifaunal species like green mussels and oysters, and partially-exposed benthic species like penshell, rocky oysters and thorny oysters. However, there are limited studies on PSP accumulation of infaunal shellfish. Initial findings showed remarkable difference in accumulation rates of the three types of shellfish. More related research is required to validate the results on other shellfish species and PSP causative organisms.

This particular study has significant importance in regulatory purposes. In cases wherein advisories and bulletins issued on shellfish safety encompassing all shellfish, there is the possibility of infaunal species being exempted. It was reported that these infaunal species might become toxic only at abnormally high *P. bahamense* var. *compressum* cell densities, Montojo et al., 2010. From the standpoint of food safety, proper risk assessment combined with implementation of regular monitoring and quality control procedures for possible market denials is necessary for the consumption of infaunal species.

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